## TMEM16A may regulate pH in the pancreatic acinar lumen

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Acidosis in the pancreatic fluid is an important component and likely a cause of pancreatitis and cystic fibrosis-related pancreatic problems, such as pancreatic insufficiency and pancreatitis. *In vitro* studies showed that the combination of protons released from acidic zymogen granules fused with the apical membrane of acinar cells during exocytosis and bicarbonate secreted from ductal cells controls pH in the pancreatic fluid (Behrendorff *et al.*, 2010; Steward *et al.*, 2005). Calcium activated chloride current (CaCC) in the apical membrane of the pancreatic acinar cell has been reported long ago (Park *et al.*, 2001). Recent study confirmed that TMEM16A accounts for the CaCC in the pancreatic acinar cell (Ousingsawat *et al.*, 2009). The permeability of TMEM16A to bicarbonate could be substantial at high free cytosolic calcium concentration (0.4 - 3μM) (Jung *et al.*, 2013); cytosolic calcium in the apical area easily reaches such concentration during physiological stimulation (Ito *et al.*, 1997). This project is aimed to prove that TMEM16A in the apical membrane of the acinar cell accounts for bicarbonate secretion into the pancreatic juice and therefore may lead a new way to restore pH in the acidified pancreatic fluid during pancreatic diseases.

**Methods.** Pancreatic acini were isolated from anaethetised c57bl/6j black mice following a previously described protocol (Behrendorff *et al.*, 2010). Video-rate 2-photon microscope with a 60× oil immersion objective was used to image exocytotic events and the acinar lumen. To image acinar lumenal pH changes we used HPTS (800 μmol/L) excited at 950 nm and fluorescence detected at 420 - 520 nm.

**Results.** Acinar lumenal pH drop and recovery in areas near exocytotic events induced by physiological stimulation were observed in 2mM HEPES buffered solution (pH = 7.4) as previously described (Behrendorff *et al.*, 2010). In 2mM $HCO_3^-$ buffered solution (pH = 7.4), a similar pH drop but faster pH recovery was observed in the acinar lumen. Inhibition of TMEM16A by a selective inhibitor, T16A(inh)-A01, slowed down the pH recovery significantly in 2mM $HCO_3^-$ buffered solution.

**Conclusion.** Bicarbonate secretion *via* TMEM16A helped pH recovery from an acid load in the acinar lumen caused by bulk proton release from exocytosis events.

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